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Laser polarimeter for measurement of optical activity of biological objects

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Abstract

In this paper has been described the polarimetric device for measurement of optical activity of biological tissues, where the source of radiation is an infrared laser with a wave λ=0.808 micron. The polarizers used are polarizing prisms of Glan – Taylor. To obtain required angular resolution (0.180/cm) has been developed a device that converts the angle of rotation of the analyzer into electrical signal, which is fed to the appropriate scan digital oscilloscope. The passage of the polarized light through the fingers of the hand was established and the angles of rotation of the polarization vector of the transmitted radiation were measured, the values of which may be determined by the content of hemoglobin in the blood.

Keywords: Polarization, biological object, optical activity, laser, blood, hemoglobin

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1. Introduction

Large number of organic compounds relates to optical activities (number of acids and esters, sugar, steroid compounds, sulfides, selenides and etc.). Many complex compounds of metals (particularly transition Ni, Co), the organometallic compounds, as well as liquid crystals have optical activity.

Particularly important is the role of optically active substances in the biosphere. Proteins, nucleic acids DNA and RNA, chlorophyll, hemoglobin and etc. also possess optical activity. Optically active are 19 out of 20 essential amino acids. Therefore, the problem of studying the optical activity of substances plays a huge role in biophysics, biochemistry and medicine.

Biological tissues are optically inhomogeneous absorbing environments with an average refractive index, greater than that of air, therefore on the segregation border bio object that is the – air, which is the part of the radiation is reflected and the rest penetrates into the tissue. Volume scattering is the cause of spread of a significant proportion of radiation in the opposite direction (backscatter) [Tuchina (2007)].

Analysis of spread polarized radiation patterns in blood supply tissues showed [Kiselev (2001)] that if you do not consider the possibility of multiple scattering, the light, that is not scattered, and transmitted without departure from its original direction should remain linearly polarized. Taking into account multiple scattering, intensity of light transmitted in the forward direction becomes larger (the tissues become more transparent), nevertheless, the transmitted light is almost completely depolarized.

The main source of light scattering in biological tissues is a difference in the refractive indices of the various components, i.e. between the mitochondria, nucleus, the other components and cell cytoplasm; or interstitial fluid and structural elements of the connective (fibrous) tissue (collagen and elastin fibers). As it is known [Tuchin (1989)], the condition of polarization of radiation, passing through the biological tissue, does not affect its absorption coefficient. Absorption is strongly affected by the temporal coherence of radiation and related monochromaticity, and hence the laser light is absorbed substantially less, than, for example, the radiation of the lamp.

2. Description of the device

First of all, it is necessary to determine the degree of radiation absorbtion, passing through the fingers. Such data is presented below. Passed radiation is determined with the use of device IMO – 2H. Laser output power comprised 105 mW.

Table 1. Attenuation of the radiation passing through the fingers.

Radiation passed	Thikness of fingers, mm	Absorbtion rate, %
Pinky – 1,4 mW	14±0,5	88,5
Ring finger – 1,1 mW	15±0,5	98,0
Middle – 1,0 mW	16±0,5	98,1
Forefinger – 0,9 mW	17±0,5	99,0

Thus, the radiation, passing through the finger is attenuated by average of 100 times.

As various biological tissues have quite different absorption spectra, the choice of laser wavelength is very important in such measurements. It is well known [Serebriakov (2010)], that the spectral range of 0.800 – 1.2 micron is the largest tissues transmissioning range. Scattering of radiation dominates at lower wavelengths. However, in this case, presence of water and glucose in biological tissues is essential. Our studies about passing through the human hand tissues laser radiation with the wavelengths λ = 0.532 micron, 0.65 micron, 0,808 micron, 0.843 micron and 1.06 micron have shown that radiation with a wavelength λ = 0.808 micron in our case is optimal.

As it is well known, for optically active pure liquids it was found by Zh.B. Bio that:

$$\varphi=[\alpha]\cdot c\cdot I \qquad (1)$$

where φ is – rotation angle of the polarization plane, $\boldsymbol{l}$ is – the thickness of the active substance, $\boldsymbol{c}$ is – the concentration of the active substance, [$\boldsymbol{\alpha}$] is – relative optical activity of the substance. It is also necessary to take note that [$\boldsymbol{\alpha}$] ~$1/\lambda^2$. This leads to the conclusion that to increase the sensitivity of polarimetric device the wavelength should be reduced but as noted above, in this case the scattering of radiation in the tissue is increasing. (Nevertheless, it should be noted that there is also anomaly, i.e. with λ close to λ_0 (λ_0– resonance absorption) *[α]* is increasing with increasing λ).

Figure 1 shows a block diagram of the polarimetric optical device for testing optical activities of biological objects. In this case fingers of human hand are used as a biological object.

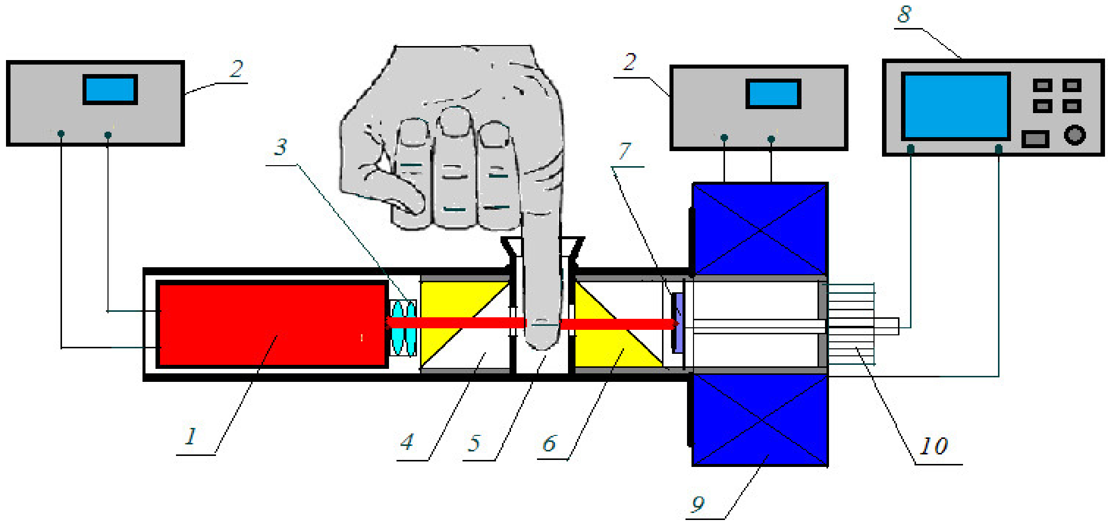

Fig.1 Block diagram of the device: 1 – semiconductor laser, 2 – power supply, 3 – optical lens, 4 – polarizer, 5 – placement of biological object, 6 –analyzer, 7 – photo detector, 8 – digital oscilloscope, 9 – converter, 10 – micrometer reel.

Semiconductor laser 1 with a wavelength λ=0.808 micron, which is powered by power supply 2 served as a radiation source. The laser beam was focused by optical lens 3. Two prisms Glan – Taylor 4 and 6 are applied as polarizers. Sensor 5 is placed between them, which houses the biological object (human hand finger). The second prism 6 serves as an analyzer. In such polarizing prisms ordinary ray passes and extraordinary ray is reflected.

The laser beam passing through the polarizer hits the object of study, such as a finger and portion of radiation a passed through it further goes to analyzer. Before the measurements, the polarizer (E_p) and the analyzer (E_a) are in a crossed state ($E_p \perp E_a$), which corresponds to a complete overlap of radiation and signal at photodetector 7 becomes minimal. With a parallel arrangement of the polarizers transmission axes ($E_p \| E_a$) signal is maximized. Then the signal from photo detector is fed to one of the inputs (axis "Y") of digital oscilloscope Tektronix 8. From the converter 9 electrical signal is fed to the other input (axis "X") of the oscilloscope. The converter is used to convert analyzer rotation angle into electrical signal, defining a scan of axis "X". Changing of the analyzer rotation angle is carried out by means of the micrometer reel 10. This device makes possible to obtain an angular resolution 0.18 °/cm, which corresponds to a single cell on digital oscilloscope. If there is an optically active object in the sensor 5, the minimum signal is shifted by an angle φ, in accordance with the formula (1).

3. Measurements results

Figure 2 shows the dependence of the photo detector signal from the rotation angle of analyzer with respect to polarizer, when there is no biological object in the sensor.

As it can be seen from the graph, the signal intensity depending on the angle of rotation of the analyzer varies according to the law of Malus (intensity of the transmitted light is proportional to $I=I_0\cos^2\varphi$). After that, object of study is placed between the polarizer and analyzer. In order to determine parameters of the device (sensitivity, accuracy in determining the angle of rotation) polarization plane rotation was measured with mica of different thickness. To do this, one of the minima was placed in the middle of the digital oscilloscope scale (curve 1 in figure 2b) and zooms in on the axis “X”. The figure shows, that increasing the thickness of the absorbing material (mica) by 5 micron, rotation angle of polarization plane is also increasing by 0.9^0, the accuracy of determining the angle of rotation depends on the accuracy of the measured minimum signal from the optically active substance. In this case, the accuracy is $\pm 0{,}2^0$.

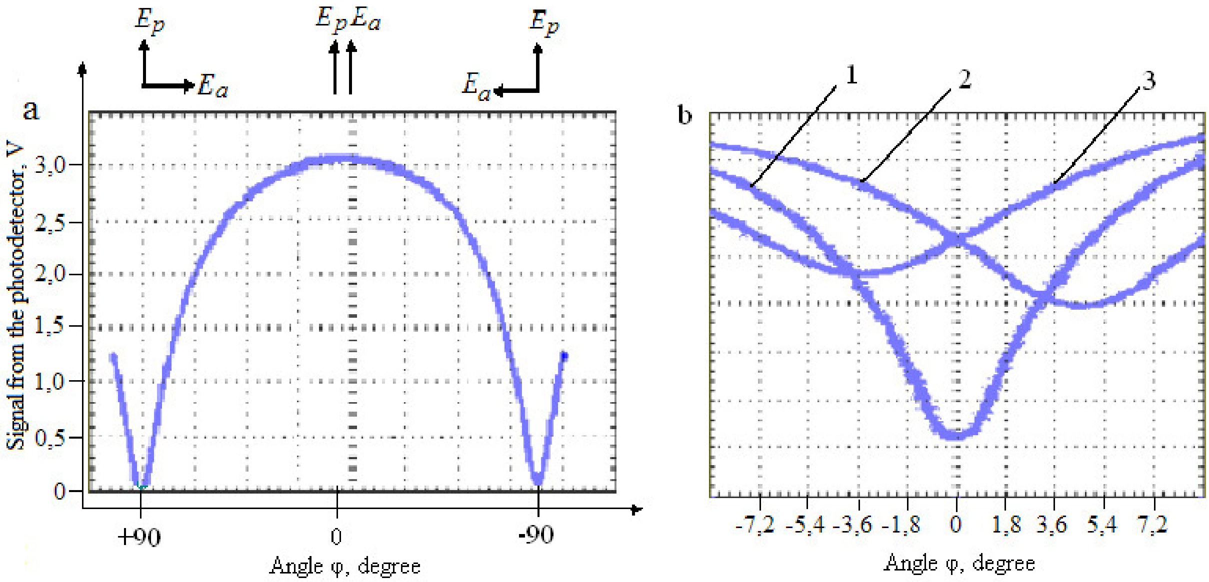

Fig. 2. (a) – dependence of photo detector signal amplitude on the analyzer rotation angle **φ** with respect to polarizer. The angular distance between the minima comprise 180^0, (b) – position of the minima of the signal, depending on analyzer rotation angle: 1 – shift of zero, 2 – 45 micron thickness mica (shift $4{,}5^0$), 3 – 40 micron thickness mica (shift $3{,}6^0$). Signal axis “Y” is equal to 500 mv/cm (one centimeter corresponds to one cell of oscilloscope screen).

In this concept of device building, oscillations of signal directly related to the movement of blood, ejected by the heart into the vessels, i.e. pulse, should be observed. It should be noted, that the frequency of the detected pulse in this case depends on the sweep speed on the axis “X”, which was defined by the speed of reel 10 rotation and varied in different experiments. However, this fact does not significantly influence on the measurements results.

Figure 3 shows the measurement of angular distribution of intensity transmitted through optically active substance (human finger) of polarized light. Figure 3 (a and b) shows the dependence of signal on the angle of analyzer for little finger (a) and forefinger (b) figure 2a scale. Figure 3 (c and d) shows the signals from little and forefinger in a scale along the axis “X” the same, as on the figure 2b. Oscillations associated with the pulse can be clearly observed.

Comparison of figures 2a and 3a,b shows that the peaks on figure 3a,b correspond to maximum transmission on figure 2a, which indicates that the polarization component of radiation passes through the thickness of fingers, however, in this scale of angular scan, it is impossible to determine the shift of minima. At a higher angular resolution (figures 3c, d) it can be seen, that signal minimum shift for little finger comprise 1^0, for forefinger 1.8^0. This difference is probably due to the fact, that forefinger has thickness slightly greater than little finger.

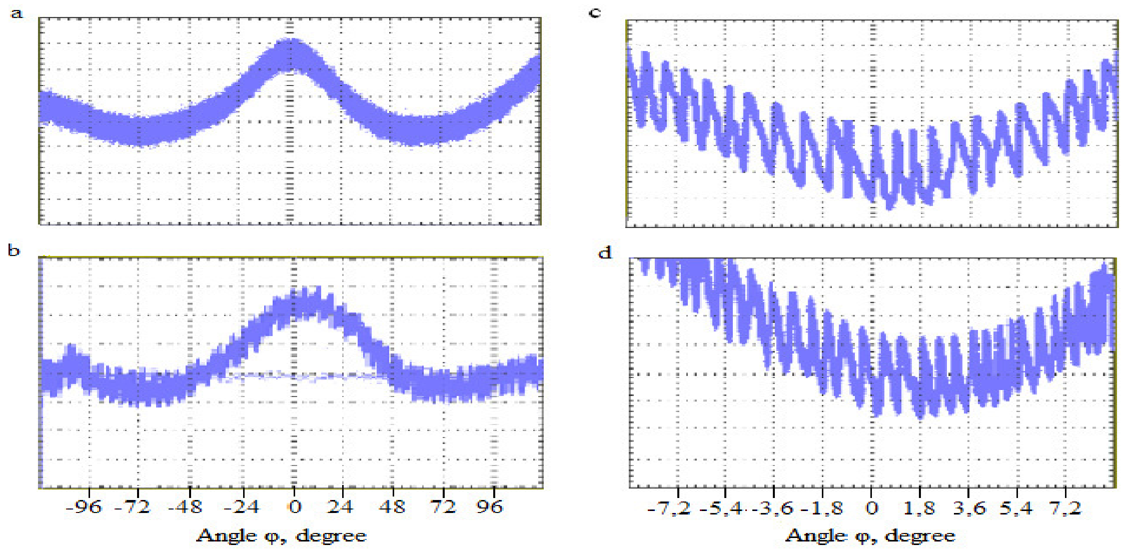

Fig. 3 The angular distribution of the signal from polarized light, transmitted through the fingers of human hand: (a) – pinky (little finger), (b) – forefinger (scan – axis "Y" – 10 mv/cm); (c) – the minimum position signal from little finger; (d) – from the forefinger (scan-axis "Y" 5 mv/cm).

Figure 4 shows a general view of the laser polarimeter.

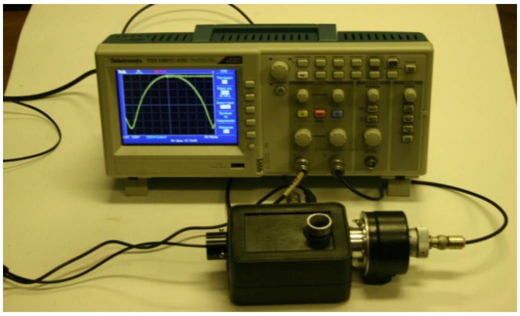

Fig. 4. General view of the device.

4. Conclusion

Assuming that optically active component in human finger is firstly blood (because of glucose and hemoglobin) the angle of rotation φ can be estimated. It is known [Beryozov (1982)], that human blood contains 130 – 160 g/l of hemoglobin. When a specific/relative rotation of hemoglobin is $+10.4^0$ [Bresler (1963)], the calculation per formulae Bio gives a value for angle of rotation in total of $\sim1^0$, which is almost identical with those of our measurements. In addition, there is an option, when polarized radiation transmitted through the biological tissue becomes elliptically polarized, and the parameters of ellipse can vary depending on the absorbing and scattering properties of the environment. Nevertheless, correspondence of experimental and calculated data requires additional studies, which will be published later. However, the latter assumption requires additional studies, which will be published later.

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